

AMENDMENTS TO THE CLAIMS

1. (Currently Amended) A multilayer composite material comprising as the inner matrix a composite material comprising:

- (i) hyaluronic acid and/or hyaluronic acid derivatives, and
- (ii) a matrix of demineralised bone and/or biocompatible and biodegradable ceramics and/or bone of autologous or allogenic or animal origin

in association with at least one layer comprising a hyaluronic acid derivative.

2. (Currently Amended) The multilayer composite material according to claim 1, wherein the hyaluronic acid in (i) is salified with organic or inorganic bases.

3. (Currently Amended) The multilayer composite material, according to any one of claims 1 and 2, wherein said hyaluronic acid derivative in (i) is selected from the group consisting of:

- A) esters of hyaluronic acid,
- B) inner esters of hyaluronic acid with a percentage,
- C) amides of hyaluronic acid,
- D) O-sulphated derivatives of hyaluronic acid,
- E) deacetylated derivatives of hyaluronic acid, and
- F) percarboxylated derivatives of hyaluronic acid.

4. (Currently Amended) The multilayer composite material according to claim 3, wherein said hyaluronic acid ester is the benzyl ester.

5. (Currently Amended) The multilayer composite material according to claim 4, wherein the benzyl ester has a degree of esterification of from 50 to 100%.

6. (Currently Amended) The multilayer composite material according to claim 5, wherein the benzyl ester has a degree of esterification of from 75 to 100%.

7. (Currently Amended) The multilayer composite material as claimed in claim 3, wherein the hyaluronic acid inner esters have an esterification degree lower than 20%.

8. (Currently Amended) The multilayer composite material as claimed in claim 7, wherein the hyaluronic acid inner esters have an esterification degree comprised between 0.05 and 5%.

9. (Currently Amended) The multilayer composite material as claimed in claim 3, wherein the amidation degree of hyaluronic acid amides (C) is lower than or equal to 15%.

10. (Currently Amended) The multilayer composite material as claimed in claim 9, wherein the amidation degree is comprised between 0, 1, and 15%.

11. (Currently Amended) The multilayer composite material as claimed ~~in claim~~ in claim 3, wherein the deacetylated hyaluronic acid has a percentage of deacetylation lower than or equal to 30%.

12. (Currently Amended) The multilayer composite material as claimed in claim 3, wherein the percarboxylated hyaluronic acid (F) has a percarboxylation degree of between 0.1 and 100%.

13. (Currently Amended) The multilayer composite material as claimed in claim 12, wherein said percarboxylation degree is comprised between 25 and 75%.

14. (Currently Amended) The multilayer composite material according to claim 1 ~~anyone of claims 1-13~~, wherein the biocompatible and biodegradable ceramics is selected from the group consisting of hydroxyapatite and/or tribasic calcium phosphate and/or calcium sulphate.

15. (Currently Amended) The multilayer composite material according to claim 1 ~~anyone of claims 1-13~~, wherein the bone matrix is partially or completely demineralised.

16. (Currently Amended) The multilayer composite material according to claim 1, ~~anyone of claims 1-15~~ wherein the hyaluronic acid derivative has a molecular weight of between 200 and 750 kDs.

17. (Currently Amended) The multilayer composite material according to claim 1, ~~anyone 1-16~~ wherein the hyaluronic acid derivative is in a form selected from the group consisting of a non woven tissue, a sponge, a paste, granules, and powders.

18. (Canceled).

19. (Currently Amended) The multilayer composite material according to claim 1, ~~18~~ wherein the layers are 2.

20. (Currently Amended) The multilayer composite material according to claim 1, ~~18~~ wherein the layers are 3.

21. (Currently Amended) The multilayer composite material, according to claim 1 ~~anyone of claims 18-20~~, wherein said hyaluronic acid derivative contained in the layer (s) is selected from the group consisting of:

- A) esters of hyaluronic acid,
- B) inner esters of hyaluronic acid with a percentage
- C) amides of hyaluronic acid,
- D) O-sulphated derivatives of hyaluronic acid,
- E) deacetylated derivatives of hyaluronic acid, and
- F) percarboxylated derivatives of hyaluronic acid.

22. (Currently Amended) The multilayer composite material according to claim 21 ~~anyone of claims 18-20~~, wherein said hyaluronic acid ester is the benzyl ester.

23. (Currently Amended) The multilayer composite material according to claim ~~20~~22, wherein the benzyl ester has a degree of esterification of from 50 to 100%.

24. (Currently Amended) The multilayer composite material according to claim 23, wherein the benzyl ester has a degree of esterification of from 75 to 100%.

25. (Original) The multilayer composite material according to claim 21, wherein the hyaluronic acid inner esters have an esterification degree lower than 20%.

26. (Currently Amended) The multilayer composite material according to claim 25, wherein the hyaluronic acid inner esters have an esterification degree comprised between 0.05 and 5%.

27. (Original) The multilayer composite material according to claim 21, wherein the amidation degree of hyaluronic acid amides (C) is lower than or equal to 15%.

28. (Original) The multilayer composite material according to claim 27, wherein the amidation degree is comprised between 0,1 and 15%.

29. (Original) The multilayer composite material according to claim 21, wherein the deacetylated hyaluronic acid has a percentage of deacetylation lower than or equal to 30%.

30. (Original) The multilayer composite material according to claim 21, wherein the percarboxylated hyaluronic acid (F) has a percarboxylation degree of between 0.1 and 100%.

31. (Currently Amended) The multilayer composite material according to claim 1 ~~claims~~ 30, wherein said percarboxylation is comprised between 25 and 75%.

32. (Currently Amended) The multilayer composite material according to claim 1 ~~anyone of claims 18-31~~, wherein the hyaluronic acid derivatives comprised in the layer(s) are in the form selected from the group consisting of: non woven material, woven material, and compact, perforated porous or microporous membranes and films.

33. (Currently Amended) The multilayer composite material according to claim 1 ~~anyone of claims 18-32~~, wherein the inner matrix is in the form of a sponge consisting of the benzyl ester of hyaluronic acid with a percentage of esterification ranging between 70 and 100%, containing inside said sponge:

- ☐ bone granules or powders that are autologous and/or allogenic and/or of animal origin, or
- ☐ granules or other two- or three-dimensional structures constituted by biodegradable ceramics or, lastly,
- ☐ partially or completely demineralised bone matrix.

34. (Currently Amended) The multilayer composite material according to claim 1 ~~133~~, subsequently coated throughout with HA and/or the derivatives thereof in the form of a thin film and/or sponge, to favour the entry, distribution, and adhesion of the cells that will migrate once they have been loaded therein.

35. (Currently Amended) The multilayer composite materials according to claim 1 ~~anyone of claims 18-34~~, wherein the inner matrix is in the form of sponges formed by the inner esters of HA containing inside them:

- ☐ bone granules and/or powders of autologous and/or allogenic type and/or of animal origin,
- ☐ biodegradable ceramics, or
- ☐ partially or completely demineralised bone matrix.

36. (Currently Amended) The multilayer composite materials according to claim 1 ~~anyone of claims 18-34~~, wherein the inner matrix is in the form granules, spheres, powders, and/or two- and three-dimensional structures of various shapes and sizes consisting of biodegradable ceramics that are coated/incorporated in a layer of HA subsequently cross-linked to form its inner ester (ACP), which thus covers all the ceramic structures.

37. (Currently Amended) The multilayer composite materials according to claim 1, ~~anyone of claims 18-34~~, wherein the inner matrix is in the form of pastes and/or gels consisting of HA derivatives enclosing bone powders and/or granules that are autologous and/or allogenic and/or of animal origin, or granules or other two- or three-dimensional structures constituted by biodegradable ceramics or, lastly, pastes and/or gels containing demineralised bone matrix.

38. (Currently Amended) The multilayer composite materials according to claim 1 ~~anyone of claims 18-34~~, wherein the inner matrix is in the form of fibres comprising the benzyl ester of HA with a percentage of esterification ranging between 50 and 100%, possibly associated with other natural polymers selected from collagen and cellulose and the derivatives thereof, or synthetic polymers selected from poly-lactic, polyglycolic and poly-caprolactone acid, in association with demineralized bone matrix and hyaluronic acid.

39. (Original) The multilayer composite materials according to claim 38, wherein the matrix can be wetted with a solution of hyaluronic acid ester, to render it more compact with the layers between which it is sandwiched.

40. (Currently Amended) The multilayer composite materials according to claim 38 ~~anyone of claims 38 and 39~~, wherein said matrix consists of fibres of hyaluronic acid benzylester having an esterification degree of 75% in amounts ranging from 10 to 50% and demineralised bone matrix in amounts ranging from 50 to 90% and hyaluronic ~~hyaluronic~~ acid having an average molecular weight ranging from 200 to 750 KDs in amounts ranging from 0.1 and 40%.

41. (Currently Amended) The multilayer composite material according to claim 40, wherein said matrix consists ~~consist~~ of fibres of hyaluronic acid benzylester having an esterification degree of 75% in amounts ranging from 14 to 24%, demineralised bone matrix in amounts varying between 60 and 80%, hyaluronic acid having an average molecular weight ranging from 500 to 700 KDs in amounts comprised between between 5 and 10%.

42. (Currently Amended) The multilayer ~~multilayer~~ composite material according to claim 1 ~~anyone of claims 18-41~~, wherein said inner matrix is immersed to make the final matrix more compact and to fixable to the layers.

43. (Currently Amended) The multilayer composite according to claim 42, wherein said polymer is selected from:

- hyaluronic acid benzyl ester with a percentage of esterification of between 55 and 100%;
- fibrin glue,
- photocross-linkable polymers, and
- collagen and derivatives thereof.

44. (Currently Amended) The multilayer composite material according to claim 1, ~~anyone of claims 18-43~~, wherein the layer(s) comprise a hyaluronic acid ester.

45. (Original) The multilayer composite materials according to claim 44, wherein said hyaluronic acid is the benzylester with a percentage of esterification ranging between 50 and 100%.

46. (Original) The multilayer composite material according to claim 45, wherein said percentage degree is comprised between 75 and 100%.

47. (Currently Amended) The multilayer composite material according to claim 44 ~~anyone of claims 44-46~~, wherein the layers are in the form of: a non-woven material, containing

fibres of the hyaluronic acid ester possibly associated with natural polymers selected from collagene and cellulose and the derivatives thereof, or synthetic polymers selected from poly-lactic acid, poly-glycolic acid, and poly-caprolactone acid.

48. (Currently Amended) The multilayer composite material according to claim 44, ~~anyone of claims 44-46~~ wherein the layers are in the form of a woven material containing fibres of the hyaluronic acid ester, possibly subsequently immersed in a solution of hyaluronic acid.

49. (Currently Amended) The multilayer composite material according to claim 1 ~~anyone of claims 44-46~~, wherein the layers are in the form of compact perforated porous or microporous membranes and films.

50. (Currently Amended) The multilayer composite materials according to claim 1, ~~anyone of claims 18-49~~ further containing pharmacologically and/or biologically active ingredients.

51. (Original) The multilayer composite materials according to claim 50, wherein the pharmacologically active ingredients are selected from the group consisting of antibiotics, antineoplastics, anti-inflammatories, cytokines, vitamins and cytotoxic, cytostatic and antiviral agents.

52. (Original) The multilayer composite materials according to claim 50, wherein biologically active ingredients contain trophic, osteoinductive, angiogenetic factors.

53. (Currently Amended) The multilayer composite material according to ~~claim 50~~52, wherein the trophic, osteoinductive, and angiogenetic factors contain BMP, TGF, PDGF, FGF, EGF, IGF₁ and VEGF.

54. (Currently Amended) The multilayer composite material according to claim 1, ~~anyone of claims 18-53~~ loaded with bone marrow cells.

55. (Currently Amended) The multilayer, composite material according to claim 1 ~~anyone of claims 18-53~~, loaded with autologous and/or allogenic mesenchymal cells either undifferentiated or partially differentiated into osteoblasts.

56. (Currently Amended) The multilayer composite materials according to claim 1 ~~anyone of claims 18-53~~, loaded with autologous and/or allogenic mesenchymal cells that are completely differentiated into osteoblasts.

57. (Currently Amended) A process for preparing the multilayer composite material according to claim 1, ~~anyone of claims 18-54~~ comprising the following steps:

- a) forming the inner matrix by associating hyaluronic acid and/or a hylauronic acid ester and demineralised bone matrix, and/or a biocompatible biodegradable ceramics and/or bone of autologous or allogenic type or of animal origin,
- b) coupling the matrix with the layer(s), and
- c) fixing the matrix to the layer(s), in toto or by means of the outer edge.

58. (Currently Amended) The process according to claim ~~54~~57, wherein step (c) is carried out by heat treatment.

59. (Currently Amended) The process according to claim 57 wherein step (c) is carried out by exposing the material coming from step (b) to a needle-punching process.

60. (Currently Amended) The process according to claim ~~58~~59, wherein step (c) is carried out by sewing the material coming from step (b) with thread made of hyaluronic acid and/or the derivatives thereof or another biocompatible and bioresorbable polymer.

61. (Currently Amended) A bone substitute or graft consisting of the multilayer composite material according to claim 1 ~~anyone of claims 1-17~~.

62. (Canceled).

63. (Currently Amended) The bone substitute or graft according to claim ~~62~~61 in the form of a sandwich or bag.

64-74. (Canceled).

75. (New) A method for forming or regenerating bone tissue, comprising surgically implanting into a patient in need thereof a bone substitute or graft consisting of a multilayer composite material comprising as the inner matrix a composite material comprising:

- (i) hyaluronic acid and/or hyaluronic acid derivatives, and
 - (ii) a matrix of demineralised bone and/or biocompatible and biodegradable ceramics and/or bone of autologous or allogenic or animal origin,
- in association with at least one layer comprising a hyaluronic acid derivative.

76. (New) The method according to claim 75 for forming or regenerating a maxillofacial bone tissue.

77. (New) The method according to claim 75 for forming or regenerating bone tissue of the shoulder, hand, and foot.

78. (New) The method according to claim 75 for forming or regenerating bone tissue in dental surgery.

79. (New) The method according to claim 75 for regenerating bone tissue degraded following to cancer.

80. (New) A method for forming or regenerating bone tissue in orthopaedic surgery comprising

- fusing in a patients in need thereof adjacent bones by means of a bone substitute or tissue graft consisting of a multilayer composite material comprising as the inner matrix a composite material comprising :
 - (i) hyaluronic acid and/or hyaluronic acid derivatives,
 - (ii) a matrix of demineralised bone and/or biocompatible and biodegradable ceramics and/or bone of autologous or allogenic or animal origin,in association with at least one layer comprising a hyaluronic acid derivative
- then forming new bone tissue.

81. (New) A method for forming or regenerating bone tissue comprising:
fusing in a patient in need thereof two adjacent vertebral bodies by means a bone substitute or graft consisting of a multilayer composite material comprising as the inner matrix a composite material comprising:

- (i) hyaluronic acid and/or hyaluronic acid derivatives, and
 - (ii) a matrix of demineralised bone and/or biocompatible and biodegradable ceramics and/or bone of autologous or allogenic or animal origin,
- in association with at least one layer comprising a hyaluronic acid derivative

82. (New) A method for forming or regenerating bone tissue comprising filling one or more vertebral bodies previously hollowed out with a bone substitute or graft consisting of a multilayer composite material comprising as the inner matrix a composite material comprising :

- (i) hyaluronic acid and/or hyaluronic acid derivatives, and
 - (ii) a matrix of demineralised bone and/or biocompatible and biodegradable ceramics and/or bone of autologous or allogenic or animal origin,
- in association with at least one layer comprising a hyaluronic acid derivative.